

Supplementary Materials: Belowground Interactions Impact the Soil Bacterial Community, Soil Fertility and Crop Yield in Maize/Peanut Intercropping Systems

Qisong Li, Jun Chen, Linkun Wu, Xiaomian Luo, Na Li, Yasir Arafat, Sheng Lin, Wenxiong Lin

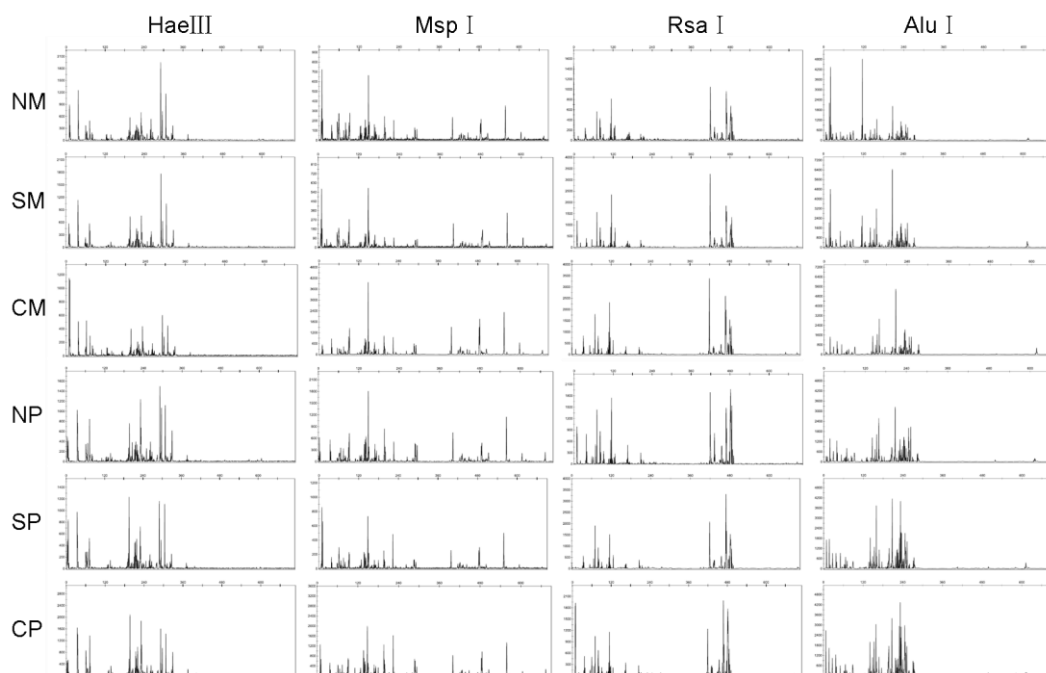


Figure S1. T-RFLP profiles of bacterial 16S rRNA genes amplified from different treatments soils for each restriction enzyme. NM: non-separated maize; SM semi-separated maize; CM: completely separated maize; NP: non-separated peanut SP: semi-separated peanut; CP: completely separated peanut.

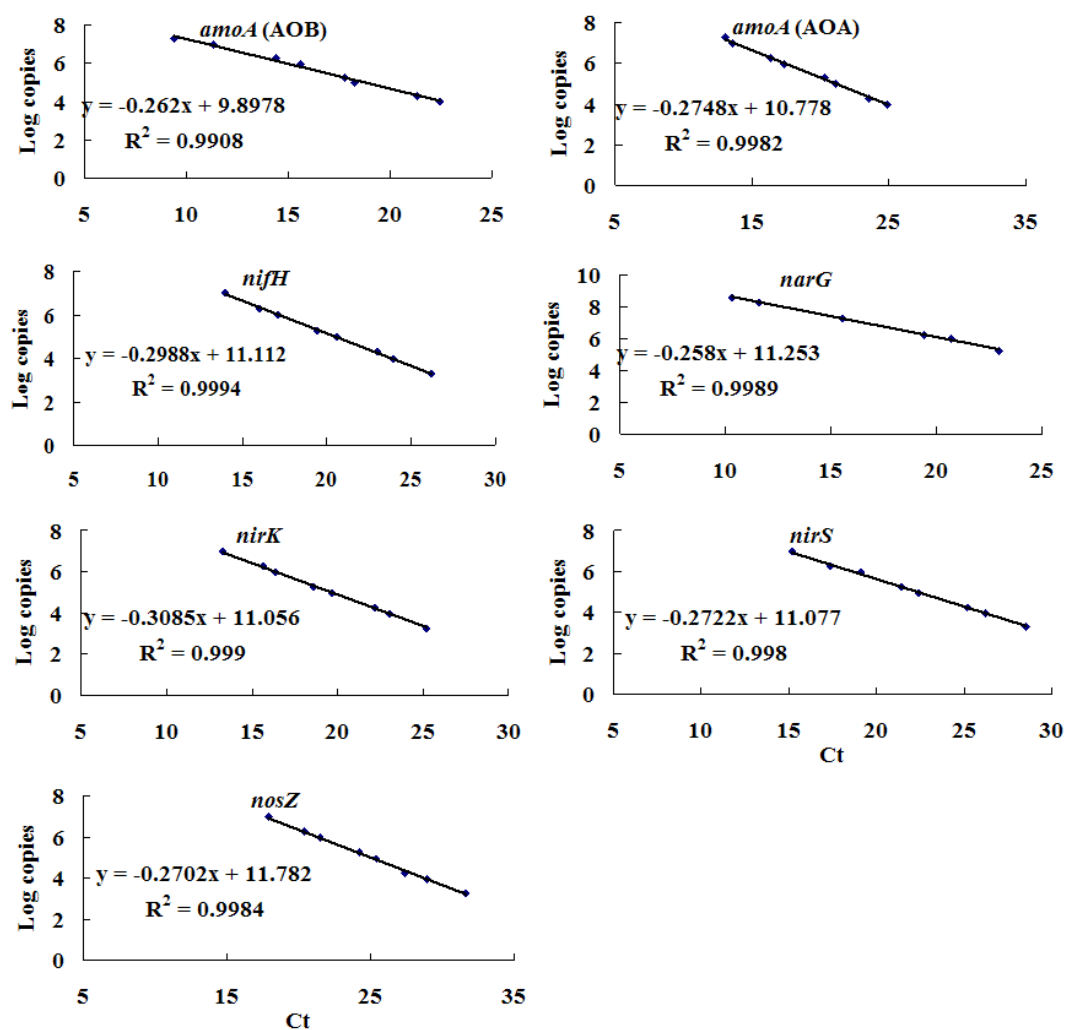


Figure S2. Standard curves for the quantification of genes involving in soil N cycling.

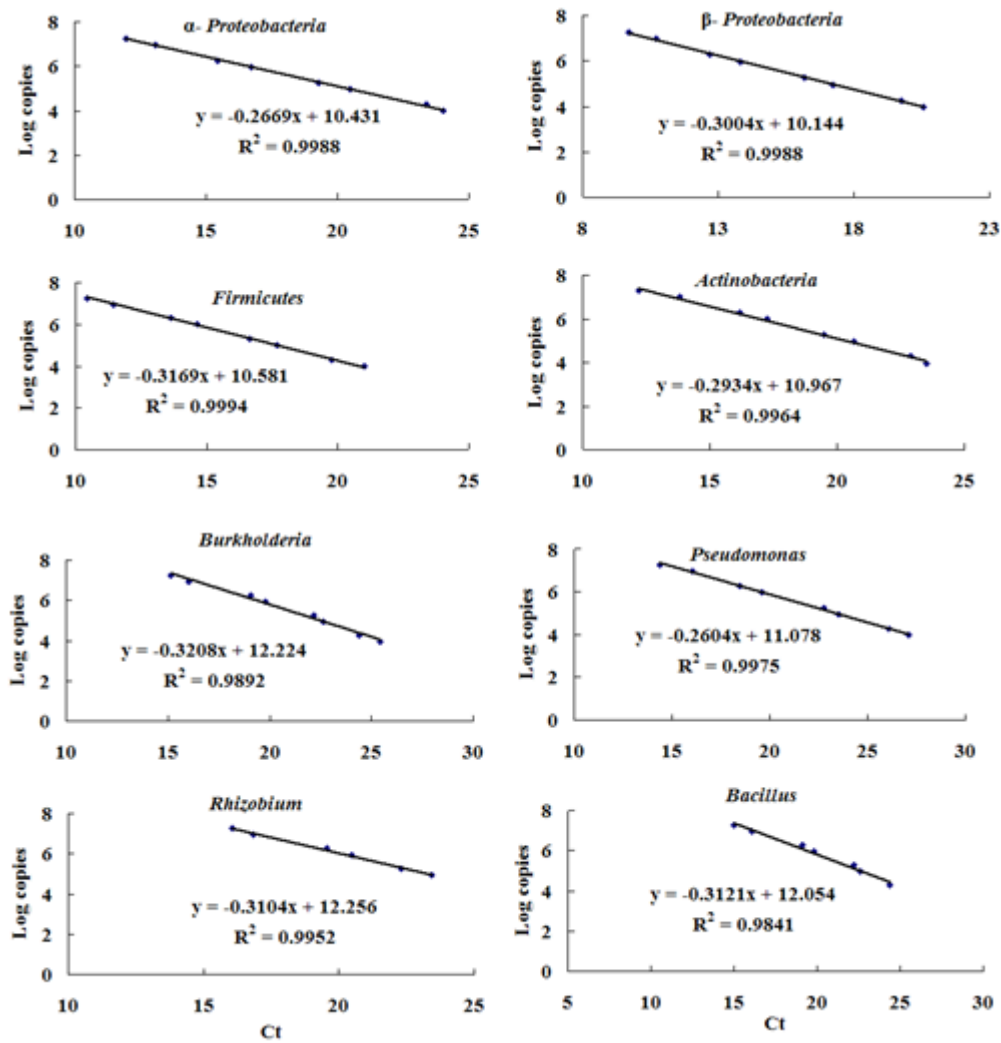


Figure S3. Standard curves for the quantification of genes which involved in major microbial communities under different intercropping patterns.

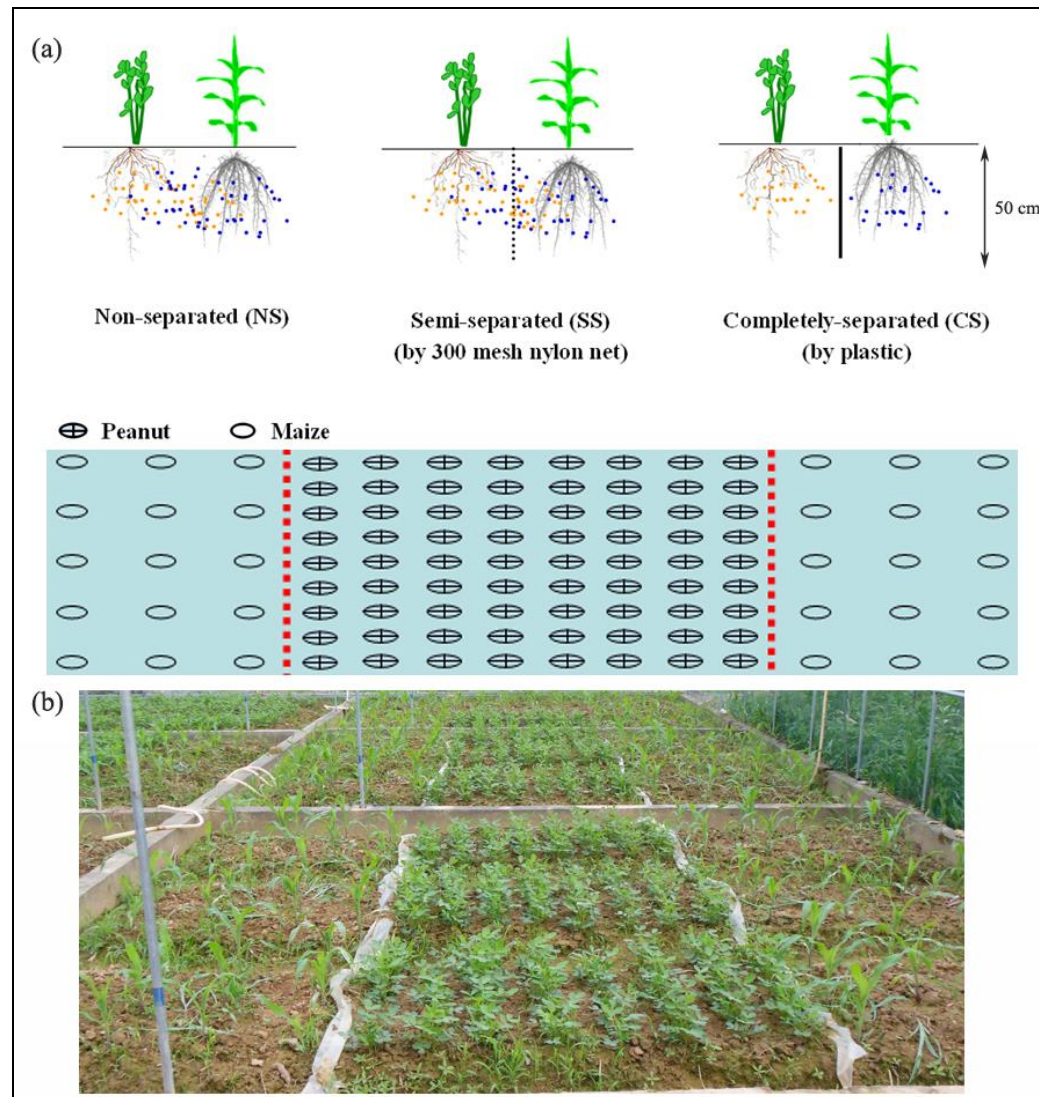


Figure S4. Field experimental designs regard maize and peanut intercropping systems. Field experimental designs (a) and actual growth status (b) under maize and peanut intercropping systems. NS: non-separation treatment; SS: semi-separation treatment; CS: completely separated treatment. Row space was 20 and 35 cm for peanut and maize treatment, respectively. Interplant space was 20 and 35 cm for peanut and maize treatment, respectively. The distance between wheat strip and maize strip was 22.5 cm. The row ratio of maize and peanut in all intercroppings were 3:8. Root barrier depth was 50 cm.

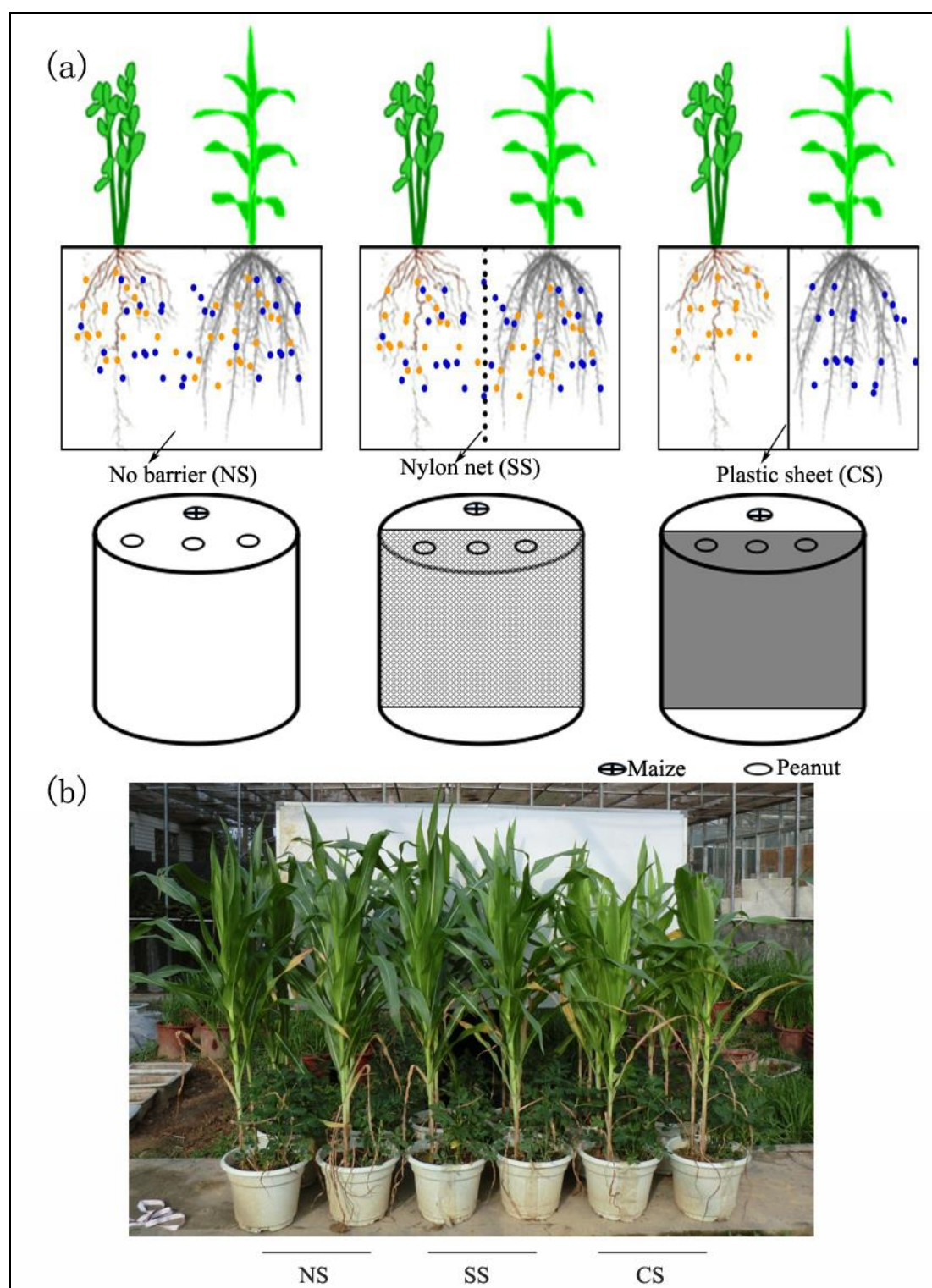


Figure S5. Pot experimental designs and actual growth status regard maize and peanut intercropping systems. Pot experimental designs (a) and actual growth status (b) under maize and peanut intercropping systems. NS: non-separation treatment; SS: semi-separation treatment; CS: complete separation treatment. Black and blue spots indicate root exudates and soil microorganisms of peanut and maize, respectively.

Table S1. Top terminal restriction fragments (T-RFs) with 20% cumulative contribution to the dissimilarity between belowground interaction intercropping (NS and SS) and completely separated intercropping (CS).

Enzymes				TRFLP-PAT assignment	Reference
<i>Msp</i> I	<i>Hae</i> III	<i>Afa</i> I	<i>Alu</i> I		
Maize					
122	219	444	234	<i>Brevibacillus brevis</i> (D78457)	[1]
150	235	894	75	<i>Paenibacillus</i> sp.	[2]
153	231	456	73	<i>Bacillus</i> sp.	[2]
171	330	459	64	clone OCS155 (AF001652)	
	123			No Match	
78	75	488	217	<i>Polyangium</i> sp.	[3]
145	309	456	73	<i>Bacillus subtilis</i> (AL009126)	[2,4]
488	217	427	148	clone T33 (Z93960)	
492	39	72	236	<i>Pseudomonas</i> sp.	[3,5]
64	63	425	144	<i>Acidosphaera</i> (D86512)	
150	227	422	250	<i>Sphingomonas</i> sp.	[6]
Peanut					
153	231	456	73	<i>Bacillus</i> sp.	[2]
487	197	562	152	<i>Burkholderia</i> sp.	[7,8]
520	222	451	237	<i>Clostridium</i> sp.	[9]
492	39	72	236	<i>Pseudomonas</i> sp.	[3,5]
139	206	451	232	str. AS2988.(AF060671)	
159	67	78	232	<i>Nocardia crassostreae</i> (U92800)	
509	218	58	67	clone Sva0556.	
294	219	444	234	<i>Brevibacillus brevis</i> (D78457)	[1]
	65			No Match	
488	217	427	148	<i>Xylophilus ampelinus</i> (AF078758)	[10]
91	282	310	73	<i>Cytophaga lytica</i> (M62796)	[11]
125	221	678	138	<i>Mesorhizobium loti</i> (D14514)	[12]
401	191	824	206	<i>Rhizobium hainanense</i> (U71078)	[13]
152	194	110	209	<i>Afipia clevelandensis</i> (M69186)	[14]
150	293	422	208	<i>Sphingomonas</i> sp. (U52146)	[6]

Table S2. PCR primers and thermal cycling conditions used for quantification of different genes.

Primers	Sequence (5' – 3')	Thermal conditions
<i>NifH</i> [15] nifH-F nifH-R	CCT ACG GGA GGC AGC AG ATT ACC GCG GCT GCT GGC A	95°C, 15 min, 1 cycle 95°C for 15 s, 60°C for 30 s, 72°C for 30 s, 80°C for 15 s, 40 cycles 95°C for 15 s, 60 to 95°C, 1 cycle
<i>NarG</i> [16] narG1960m2F narG2050m2R	TAY GTS GGG CAG GAR AAA CTG CGT AGA AGA AGC TGG TGC TGT T	95°C, 15 min, 1 cycle 95°C for 15 s, 65 to 60°C for 30 s (-1°C by cycle), 72°C for 30 s, 80°C for 15 s, 6 cycles 95°C for 15 s, 60°C for 30 s, 72°C for 30 s, 80°C for 15 s, 40 cycles 95°C for 15 s, 60 to 95°C, 1 cycle

<i>NirK</i> [17] nirK876 nirK1040	ATY GGC GGV CAY GGC GA GCC TCG ATC AGR TTR TGG TT	95°C, 15 min, 1 cycle 95°C for 15 s, 63 to 58°C for 30 s (-1°C by cycle), 72°C for 30 s, 80°C for 15 s, 6 cycles 95°C for 15 s, 60°C for 30 s, 72°C for 30 s, 80°C for 15 s, 40 cycles 95°C for 15 s, 60 to 95°C, 1 cycle 95°C, 15 min, 1 cycle
<i>nirS</i> [18] nirSCd3aFm nirSR3cdm	AAC GYS AAG GAR ACS GG GAS TTC GGR TGS GTC TTS AYG AA	95°C for 15 s, 65 to 60°C for 30 s (-1°C by cycle), 72°C for 30 s, 80°C for 15 s, 6 cycles 95°C for 15 s, 60°C for 30 s, 72°C for 30 s, 80°C for 15 s, 40 cycles 95°C for 15 s, 60 to 95°C, 1 cycle 95°C, 15 min, 1 cycle
<i>NosZ</i> [18] nosZ2F nosZ2R'	CGC RAC GGC AAS AAG GTS MSS GT CAK RTG CAK SGC RTG GCA GAA	95°C for 15 s, 65 to 60°C for 30 s (-1°C by cycle), 72°C for 30 s, 80°C for 15 s, 6 cycles 95°C for 15 s, 60°C for 30 s, 72°C for 30 s, 80°C for 15 s, 40 cycles 95°C for 15 s, 60 to 95°C, 1 cycle 95°C, 10 min, 1 cycle
<i>amoA</i> (AOB) [19] amoA-1F amoA-2R	GGG GTT TCT ACT GGT GGT CCC CTC KGS AAA GCC TTC TTC	94°C for 45 s, 58°C for 45 s, 72°C for 45 s, 39 cycles 95°C for 15 s, 60°C for 30 s, to 95°C for 15 s, 1 cycle
<i>amoA</i> (AOA) [20] 19F CrenamoA616r48x	ATG GTC TGG CTW AGA CG GCC ATC CAB CKR TAN GTC CA	95°C, 10 min, 1 cycle 94°C for 45 s, 55°C for 45 s, 72°C for 45 s, 39 cycles 95°C for 15 s, 60°C for 30 s, to 95°C for 15 s, 1 cycle
Actinobacteria [21] Actino235 Eub518	CGC GGC CTA TCA GCT TGT TG ATT ACC GCG GCT GCT GG	95°C, 15 min, 1 cycle 95°C for 1 min, 60°C for 30 s, 72°C for 1 min, 40 cycles
Alphaproteobacteria [21] Eub338 Alfa685	ACT CCT ACG GGA GGC AGC AG TCT ACG RAT TTC ACC YC TAC	95°C, 15 min, 1 cycle 95°C for 1 min, 60°C for 30 s, 72°C for 1 min, 40 cycles
Betaproteobacteria [21] Eub338 Bet680	ACT CCT ACG GGA GGC AGC AG TCA CTG CTA CAC GYG	95°C, 15 min, 1 cycle 95°C for 1 min, 60°C for 30 s, 72°C for 1 min, 40 cycles
Firmicutes [21] Lgc353 Eub518	GCA GTA GGG AAT CTT CCG ATT ACC GCG GCT	95°C, 15 min, 1 cycle 95°C for 1 min, 60°C for 30 s, 72°C for 1 min, 40 cycles

GCT GG

<i>Pseudomonas</i> sp. [21]	TTA GCT CCA CCT	95°C, 15 min, 1 cycle
PsF	CGC GGC	95°C for 1 min, 64°C for 30 s, 72°C for 1
PsR	GGT CTG AGA GGA	min, 40 cycles
	TGA TCA GT	
<i>Burkholderia</i> sp. [21]	CTG CGA AAG CCG	95°C, 15 min, 1 cycle
Burk3	GAT	95°C for 1 min, 64°C for 30 s, 72°C for 1
BurkR	TGC CAT ACT CTA	min, 40 cycles
	GCY YGC	
<i>Bacillus</i> sp. [21]	GGG AAA CCG GGG	95°C, 15 min, 1 cycle
BacF	CTA ATA CCG GAT	95°C for 1 min, 63°C for 30 s, 72°C for 1
1378	CGG TGT GTA CAA	min, 40 cycles
	GGC CCG GGA ACG	
<i>Rhizobium</i> sp. [22]	CCC GGC TAC YTG	94°C, 15 min, 1 cycle
F979	CAG AGA TG	94°C for 20 s, 60 °C for 30 s, 72°C for 30 s,
R1264	TAG CTC ACA CTC	40 cycle
	GCG TGC TC	

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